

A New Species of *Doriopsilla* (Nudibranchia: Dendrodorididae) from the Pacific Coast of North America, Including a Comparison with *Doriopsilla albopunctata* (Cooper, 1863)

TERRENCE M. GOSLINER

Department of Invertebrate Zoology and Geology, California Academy of Sciences, Golden Gate Park, San Francisco, California 94118, USA

MARIA C. SCHAEFER

Department of Biology, San Francisco State University 1600 Holloway Avenue, San Francisco, California 94132, USA

AND

SANDRA V. MILLEN

Department of Zoology, University of British Columbia 6270 University Boulevard, Vancouver, B.C., Canada V6T 1Z4

Abstract. Much confusion has surrounded the systematics of the yellow species of Dendrodorididae inhabiting the Pacific coast of North America. Re-examination of *Doris albopunctata* Cooper, 1863, indicates that it is properly placed in *Doriopsilla*. Examination of specimens from different localities throughout California, the Pacific coast of Baja California, and within the Gulf of California, indicates that all white-gilled specimens are conspecific with *D. albopunctata*, and that *D. fulva* (MacFarland, 1905) and *D. reticulata* (Cockerell & Eliot, 1905) are regarded as junior synonyms. A second species with yellow gills is here described as *Doriopsilla gemela*. It differs from *D. albopunctata* in aspects of its color pattern, external morphology, digestive system, reproductive anatomy, and developmental biology. The two species also differ in allozyme allelic frequencies. *Doriopsilla gemela* and *D. albopunctata* are also compared to other members of the genus present in the eastern Pacific. These comparisons show that, while *D. gemela* and *D. albopunctata* are externally similar to each other, their internal anatomy is more similar to other species than to each other.

INTRODUCTION

Much confusion has surrounded the systematic status of yellow porostomate dorids on the Pacific coast of North America. Cooper (1863) described *Doris albopunctata* from Santa Barbara, California. MacFarland (1905) later described *Doriopsis fulva* from Monterey Bay, California, and Cockerell & Eliot (1905) described *Doridopsis reticulata* from San Pedro, California. Steinberg (1961), Röllner (1970), McDonald (1983), and Valdés & Ortea (1997) considered these three species as synonymous. Behrens (1980, 1991) considered *Dendrodoris fulva* as distinct from *Doriopsilla albopunctata*. Behrens (1980, 1991), McDonald & Nybakken (1980), and McDonald (1983) considered another species (as *Dendrodoris* sp. 1 and *Dendrodoris* sp. A, respectively) with yellow rather than white gills and different egg ribbon shape as a distinct, undescribed species. Despite the above differences, no detailed examination of anatomy, developmental biology, or genetic differences has been undertaken. This paper

examines these taxa in detail to determine systematic relationships.

MATERIALS AND METHODS

In order to study the anatomy, developmental biology, and genetic variability of the two species of *Doriopsilla*, numerous specimens of both species were collected and examined. Specimens for anatomical study were collected from five localities along the central and southern California coast and from several localities on both coasts of Baja California. More than 15 individuals of each species were examined to ascertain intraspecific and interspecific anatomical variability.

For developmental studies, live specimens of *Doriopsilla albopunctata* were collected from five localities along the central and southern California coast. Specimens of *D. gemela* were collected from Hill Street, San Diego. Specimens were maintained in the seawater system at San Francisco State University and at the Steinhart Aquarium of the California Academy of Sciences.

Allozyme samples were conducted on 54 nudibranchs. Of these individuals 12 were *Doriopsilla gemela* from Hill Street. Forty-two individuals of *D. albopunctata* were sampled for allozyme comparison, including 12 specimens from Hill Street, San Diego, eight from Bird Rock, San Diego, eight from Diablo Canyon, San Luis Obispo County, 11 from Carmel Point, Monterey County, and three from Pillar Point, San Mateo County. Specimens were dissected. The anterior half of each animal was fixed in Bouin's fixative for anatomical comparison as vouchers. The posterior portions of the fresh tissue samples were homogenized in 1:1 volume ratio of tissue to homogenizing buffer. All samples were blotted on Whatman #2 filter paper. Excess moisture was wiped off of samples, and samples were placed in horizontal 11% starch gels. Gels were cooled by placing Blue Ice packs on top of the running gel. Two gel conditions were run at 150 volts on Heathkit model IP-17 power supplies. One gel system was Poulik's discontinuous buffer (electrode buffer pH 8.2, gel buffer pH 8.7), and was run for 5 hours. The other gel system was an amine citrate continuous buffer system at pH 7.8, and was run for 3.5 hours. Staining of the gels was undertaken by standard procedures. Ten allozyme stains were attempted. The allozymes attempted on the Poulik's gel were Superoxide dismutase (S.O.D.), Tri-Peptidase-1 and -2 (Trip-1, Trip-2), Phosphoglucose Isomerase (P.G.I.), Mannose Phosphate Isomerase (M.P.I.), and Phosphoglucomutase (P.G.M.). The allozymes attempted on the Amine-citrate gel were Superoxide dismutase (S.O.D.), Creatin Kinase (C.K.), Adenalin Kinase (A.K.), Malate Dehydrogenase (M.D.H.), Isocitrate dehydrogenase (I.D.H.), and 6 Phosphogluconic Acid Dehydrogenase (6P.G.D.H.). These allozymes were chosen because they have a high success rate of staining in a broad array of animal species, and several overlapped with those used in previous nudibranch allozyme investigations (Havenhand et al., 1986 and Morrow et al., 1992). The six allozymes which were scored under these gel conditions were Superoxide Dismutase, Tri-Peptidase 1 and 2, Malate Dehydrogenase, Phosphoglucose Isomerase, and Phosphoglucomutase.

SPECIES DESCRIPTIONS

Doriopsilla albopunctata (Cooper, 1863)
(Figures 1A,B, 2A,D, 3A,B, 4A)

Doris albopunctata Cooper 1863: 58.

Doriopsis reticulata Cockerell in Cockerell & Eliot, 1905: 41-42, pl. 7, fig. 5.

Doriopsis fulva MacFarland, 1905: 45.

(see McDonald, 1983 for complete synonymy)

Distribution: Known from Puerto Peñasco and Bahía de los Angeles, Gulf of California, México; Bahía Tortugas, Baja California Sur, México to Van Damme State Beach, Mendocino County, California (Marcus & Marcus, 1967; Behrens, 1991; present study).

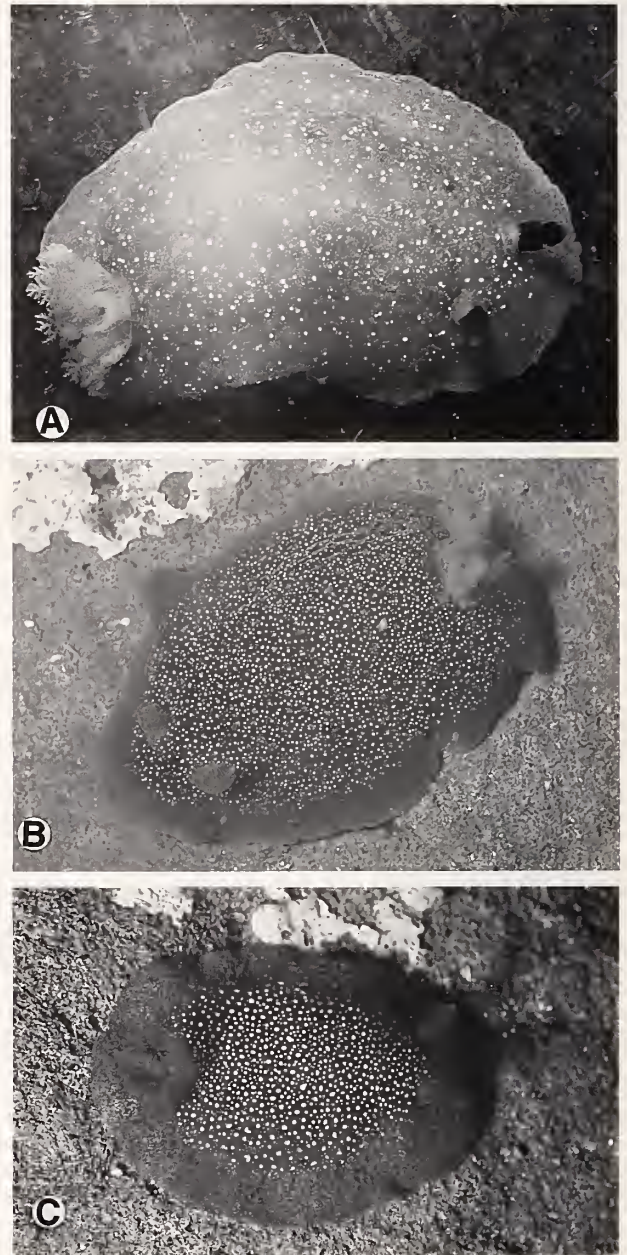


Figure 1

Living animals. A. *Doriopsilla albopunctata* (Cooper, 1863), specimen from Monterey, California showing low dorsal spot density. B. *Doriopsilla albopunctata* (Cooper, 1863), specimen from San Diego, California, showing high dorsal spot density. C. *Doriopsilla gemela* Gosliner, Schaefer & Millen, sp. nov., specimen from San Diego, California.

Material examined: One specimen, dissected, CASIZ 111388, intertidal zone, Carmel Point, Monterey County, California, 29 July 1996, M. Schaefer, coll.. One specimen, dissected, CASIZ 111389, intertidal zone, Pillar Point, San Mateo County, California, 3 June 1996, T.

Gosliner, coll. One specimen, dissected, CASIZ 111390, Hill Street, San Diego, California, 1 August 1996, M. Schaefer, coll. One specimen, dissected, CASIZ 111391, Punta Gringa, Bahía de los Angeles, Baja California, México, March 1997, H. Bertsch, coll. One specimen, dissected, CASIZ 112215, Punta Gringa, Bahía de los Angeles, Baja California, México, March 1997, H. Bertsch, coll. Two specimens dissected, intertidal zone, Pacific Grove, Monterey County, California, June 1976, S. Millen, coll. Two specimens dissected, 23–27 m depth, Scripps Canyon, La Jolla, California, 1 September 1996, M. Miller, coll. Two specimens, CASIZ 071498, intertidal zone, Centro de Acuacultura, Bahía Tortugas, Baja California Sur, México, 29 June 1984, T. Gosliner, coll. Six specimens, one dissected, CASIZ 072134, 20–23 m depth, Roca Ben, Baja California, México, 20 August 1987, R. Van Syoc, T. Gosliner, coll.

External morphology: The living animals (Figure 1A,B) reach a maximum of 60 mm in length. The body color ranges from bright yellow, orange to chestnut brown. The dorsal surface is ornamented with opaque white spots, some of which are present in the center of conical tubercles. The tubercles are 0.6–1.0 mm in diameter. The spots in the center of the tubercles are small glands, which are bordered by spicules. The size and density of tubercles varies greatly, within and between localities. Specimens from northern California are usually bright yellow throughout, but may occasionally have a central patch of chestnut brown on the dorsum. Specimens from southern California exhibit much more variation in color than do northern California specimens, but are generally much darker in color. The rhinophores are orange-yellow to yellow, with 11–30 lamellae. There are five to six bi- or tripinnate gills which are white to pale yellow in color. The notum is densely spiculate. The foot is elongate, but is generally completely covered by the posterior end of the mantle. The head is poorly developed with minute ridges.

Internal morphology: The oral tube (the presumed homolog of the buccal mass in other dorids that possess a radula) lacks any vestige of a radula. It (Figure 2A) is elongate and tubular and can be wider posteriorly when the tube is not fully extended. It passes through the anterior nerve ring, and forms a junction with the esophagus. Posteriorly, the esophagus is narrower than the oral tube and is uniformly cylindrical and glandular. From the junction of the oral tube and the esophagus, immediately anterior to the nerve ring, a pair of muscles emerges and joins the nerve ring. A second pair of muscles attaches to the base of the buccal ganglia and traverses the length of the glandular portion of the esophagus and joins the muscular portion of the esophagus (gizzard of Marcus & Marcus, 1967). These muscles function as retractor muscles for the oral tube. Posterior to the short muscular section of the esophagus is another glandular segment which

enters the stomach within the bilobed, highly digitate digestive gland. At the junction of the short muscular portion of the esophagus and this glandular segment, a second pair of retractor muscles emerges and connects posteriorly with connective tissue near the gills. The intestine emerges between the two lobes of the digestive gland. Here the widest portion of the intestine has several laterally directed glandular lobes. At this point, a short rounded pyloric caecum extends from the intestine, where it is situated ventrally. The intestine narrows and continues posteriorly to the anus, which is situated to the far left side, between the left lateral branchial plumes.

The reproductive system (Figure 2D) is triaulic. A short preampullary duct widens into an elongate, cylindrical ampulla. The ampulla divides into a short oviduct, which enters the female gland mass and the more elongate vas deferens. The proximal portion of the vas deferens expands into a wide, flattened, lobed prostatic portion which envelops most of the bursa copulatrix. From the distal end of the prostatic portion, the vas deferens narrows abruptly, then gradually widens and curves into the penial bulb. The penial bulb lacks a distinct penial papilla and contains approximately 16 rows of curved, acutely pointed penial hooks which are approximately 25–35 μm wide at the base and up to 50 μm in length (Figure 3A,B). The vaginal opening, like the penial bulb, is also narrow. The vagina is relatively narrow and straight. At its proximal end is a large, thin-walled, spherical bursa copulatrix. The slender receptaculum seminis is extremely elongate and extends beyond the proximal end of the bursa copulatrix. The duct of the receptaculum seminis joins the vagina proximally, near the base of the bursa copulatrix. Near this junction, the uterine duct emerges and joins the female gland mass. The female gland mass is large and completely developed in all specimens examined.

Developmental biology: Egg ribbons (Figure 4A) are in the form of a long, narrow, spiral ribbon attached on one edge, consisting of one to three whorls. The ribbon is crenulate along its free edge and may be 2–4 mm in height. This ribbon shape is classified as type A (Todd, 1983). Egg laying in Monterey Bay occurs throughout the year, with an increase in the summer months (MacFarland, 1906). The ribbon size, height, and number of whorls are variable, and are dependent on the size of the adult which produced it. Egg ribbon color can be yellow, orange, or off-white. These ribbons almost always have a single egg per capsule. The occurrence of two larvae per egg is rare. Capsules vary from 180 to 240 μm across, with larvae initially measuring 100–150 μm across. Planktotrophic larvae with type B shells (Todd, 1981), previously known as type 1 (Thompson, 1961) hatch after 31 days at 14°C.

Genetic variation: To ascertain genetic variability within and between populations of *Doriopsilla albopunctata*, 12

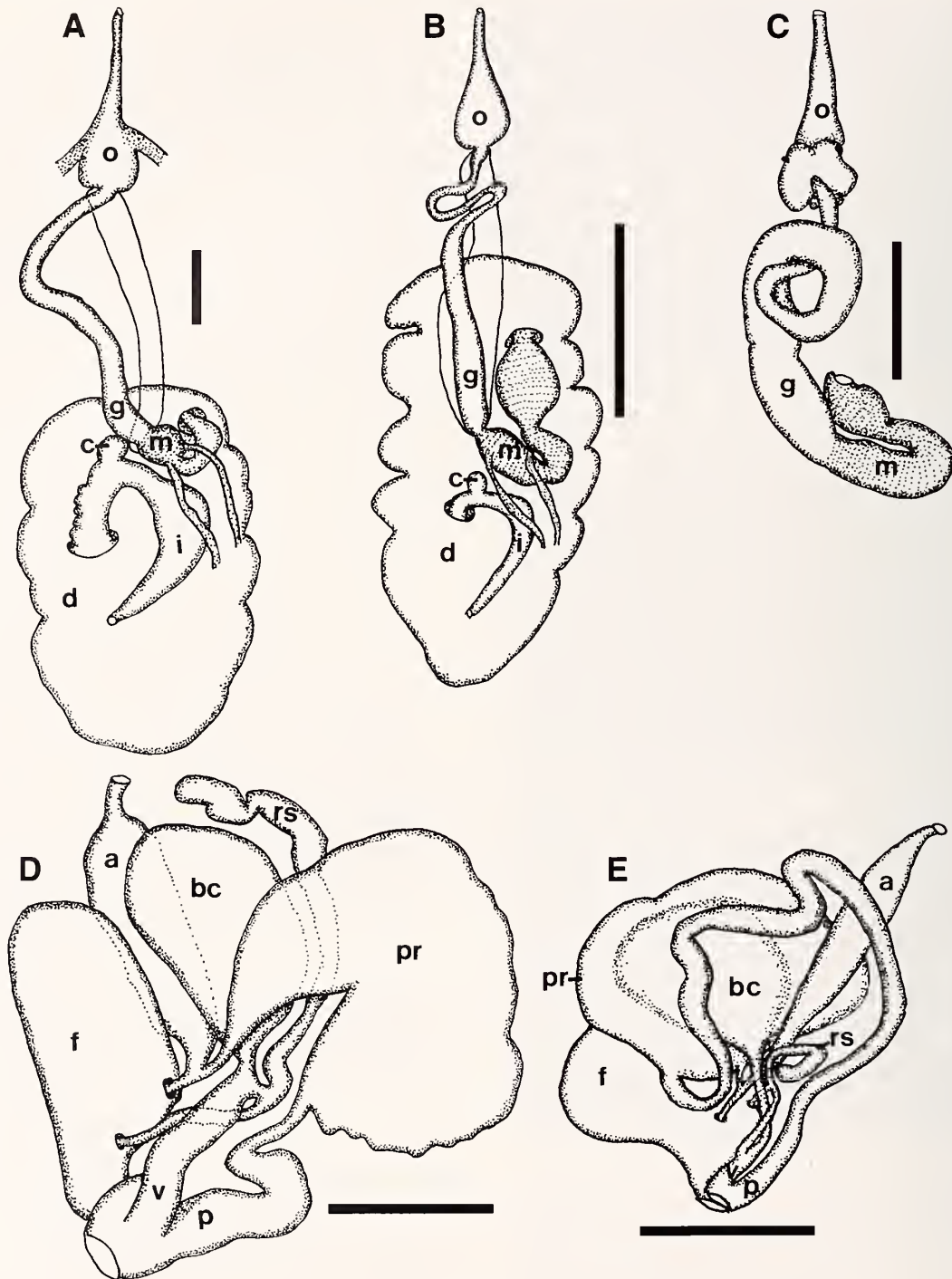


Figure 2

A. *Doriopsilla albopunctata* (Cooper, 1863), oral tube and esophagus of specimen from Bahía de los Angeles (CASIZ 112215), with blood gland and central nervous system removed, c = caecum; d = digestive gland; g = glandular portion of esophagus; i = intestine; m = muscular portion of esophagus; o = oral tube, scale = 3 mm.

B. *Doriopsilla gemela* Gosliner, Schaefer & Millen, sp. nov., oral tube and esophagus of specimen from Bahía Tortugas (CASIZ 071505), with blood gland and central nervous system removed, c = caecum; d = digestive gland; g = glandular portion of esophagus; i = intestine; m = muscular portion of esophagus; o = oral tube, scale

individuals from Hill Street, San Diego County were compared with 11 individuals from Carmel Point, Monterey County; the two localities are 685 km apart. Six loci were examined for allozymatic variation (Schaefer, in preparation). The genetic identity measure, I (Nei, 1972), within this species for the two different populations is 0.950, using allozyme comparisons. The I value ranges from 1 (if the populations are identical) to 0 (no common alleles). Conspecific populations have I values above 0.9 among varied plants and animals (Thorpe, 1983; Nei, 1987). The genetic distance, D (Nei, 1972), was 0.052 between the two populations. Values for D vary from 0 (if the populations are identical) to infinity (no common alleles). Allelic frequencies at both sites were within expected parameters as to indicate Hardy-Weinberg Equilibrium, as determined by chi square analysis.

Doriopsilla gemela Gosliner, Schaefer & Millen,
sp. nov.

(Figures 1C, 2B,C,E, 3C–E, 4B)

yellow-gilled porostome Behrens, 1980:102, fig. 146.

Dendrodoris sp. A McDonald & Nybakken, 1980:54–55, fig. 57; McDonald, 1983:171.

Dendrodoris sp. 1 Behrens, 1991:71, fig. 130.

Distribution: Known from the Gulf of California, México, from Bahía de los Angeles and along the Pacific coast of North America from Bahía Tortugas, Baja California Sur, México to Elkhorn Slough, Monterey County, California (Behrens, 1991; present study).

Etymology: The name *gemela* comes from the Spanish for twin, as this species is externally similar to its sympatric congener *Doriopsilla albopunctata*.

Type material examined: Holotype, CASIZ 111392, intertidal zone, Hill Street, San Diego, California, 1 August 1996, M. Schaefer, coll. Paratypes: One specimen, CASIZ 111393, same locality, date, and collector as holotype. One specimen, CASIZ 111394, same locality, date, and collector as holotype. One specimen, dissected, CASIZ 111395, same locality, date, and collector as holotype. Fifteen specimens, one dissected, CASIZ 071505, intertidal zone, Centro de Acuicultura, Bahía Tortugas, Baja California Sur, México, 29 June 1984, T. Gosliner, et al., coll. Three specimens, one dissected, CASIZ 074648, in-

tertidal zone, Centro de Acuicultura, Bahía Tortugas, Baja California Sur, México, 28 June 1984, S. Klontz, D. Catania, and R. Van Syoc, coll. One specimen, CASIZ 074649, 3–5 m depth, Los Morros, mouth of Bahía Tortugas, Baja California Sur, México, 1 July 1984, T. Gosliner, coll. One specimen, CASIZ 074647, intertidal zone, Centro de Acuicultura, Bahía Tortugas, Baja California Sur, México, 1 July 1984, T. Gosliner, coll. One specimen, CASIZ 073523, 7 m. depth, Punta Gringa, Bahía de los Angeles, Baja California México, 20 September 1991, T. Gosliner, coll. Seven specimens, two dissected, CASIZ 074642, intertidal zone, Centro de Acuicultura, Bahía Tortugas, Baja California Sur, México, 2 July 1984, H. Bertsch, coll. One specimen, CASIZ, 071661, 9 m. depth, Punta Gringa, Bahía de los Angeles, Baja California México, 30 June 1987, S. Millen, coll.

External morphology: The living animals (Figure 1C) reach a maximum of 40 mm in length. The body color is bright yellow to orange or orange-brown. The dorsal surface appears smooth but has some minute tubercles, 0.20–0.24 mm in diameter. The notum is ornamented with small opaque white spots. The rhinophores are orange-yellow to yellow, with 7–10 lamellae. There are five to seven bi- or tripinnate gills, which are bright yellow to orange in color. The notum is densely spiculate. The foot is elongate, but is generally completely covered by the posterior end of the mantle. The head is poorly developed with triangular, furrowed tentacles.

Internal morphology: The buccal mass lacks any vestige of a radula. The oral tube (Figure 2B, C) is elongate and tubular. It is widest posteriorly at its junction with the narrow, glandular esophagus. The esophagus narrows at the point where it passes through the anterior nerve ring. Posteriorly it is cylindrical, and widens gradually forming one or more loops. The granular surface appears to contain glandular cells. Posterior to the elongate glandular segment is an elongate, curved muscular portion of the esophagus, which widens into a short, rounded glandular section immediately anterior to where it joins the stomach within the bilobed digestive gland. The two lobes of the digestive gland are well separated from each other, and their outer edges are partially subdivided by vertical partitions in the body wall. The intestine emerges between the two lobes of the digestive gland. At this point a short rounded pyloric caecum extends dorsally from the intes-

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= 2 mm. C. *Doriopsilla gemela* Gosliner, Schaefer & Millen, sp. nov., oral tube and esophagus of specimen from Hill Street, San Diego (CASIZ 111395) with blood gland removed g = glandular m = muscular portion of esophagus; o = oral tube, scale = 1 mm. D. *Doriopsilla albopunctata* (Cooper, 1863), reproductive system of specimen from Pillar Pt. (CASIZ 111389), a = ampulla; bc = bursa copulatrix; f = female gland mass; p = penis; pr = prostate; rs = receptaculum seminis; v = vagina, scale = 2 mm. E. *Doriopsilla gemela* Gosliner, Schaefer & Millen, sp. nov., reproductive system of specimen from Hill Street, San Diego (CASIZ 111395), a = ampulla; bc = bursa copulatrix; f = female gland mass; p = penis; pr = prostate; rs = receptaculum seminis; v = vagina, scale = 1 mm.

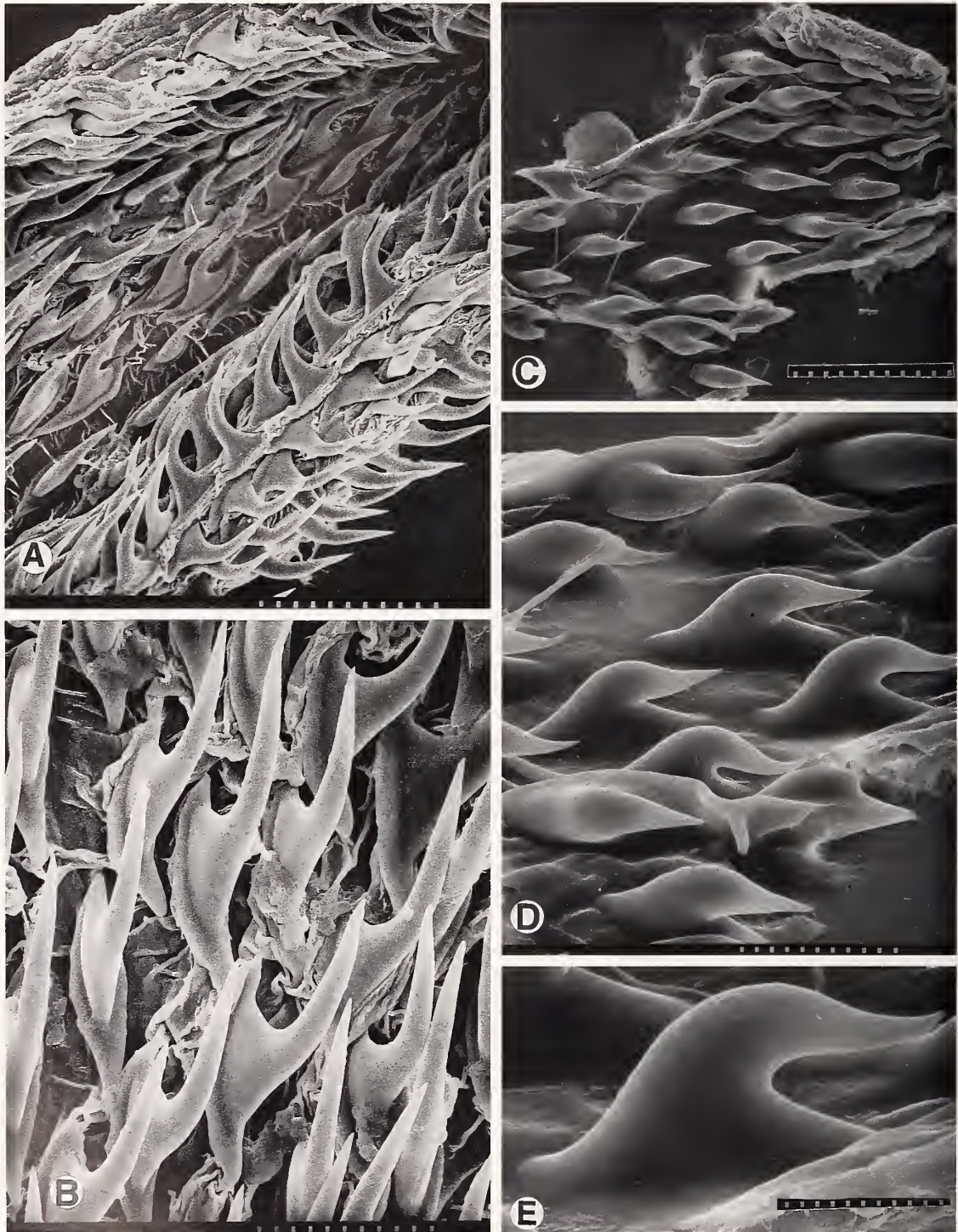


Figure 3

Penial spines. A. *Doriopsilla albopunctata* (Cooper, 1863), from Pillar Point, San Mateo County, (CASIZ, 111389), showing entire width of vas deferens, scale = 75 μm . B. *Doriopsilla albopunctata* (Cooper, 1863), isolated spines, scale = 30 μm . C. *Doriopsilla gemela* Gosliner, Schaefer & Millen, sp. nov., from Bahía Tortugas (CASIZ 071505), entire width of vas deferens, scale = 30 μm . D. *Doriopsilla gemela* Gosliner, Schaefer & Millen sp. nov., from Bahía Tortugas (CASIZ 071505), vas deferens, scale = 15 μm . E. *Doriopsilla gemela* Gosliner, Schaefer & Millen, sp. nov., from Bahía Tortugas (CASIZ 071505), isolated penial spine, scale = 7.5 μm .



Figure 4

Egg masses. A. *Doriopsilla albopunctata* (Cooper, 1863), from Carmel Point, California. B. *Doriopsilla gemela* Gosliner, Schaefer & Millen, sp. nov., from Hill Street, San Diego, California.

tine. The intestine narrows and continues posteriorly to the anus, which is on the left side, situated between the left lateral branchial plumes.

The reproductive system (Figure 2E) is triaulic. A short preampullary duct widens into an elongate, cylindrical ampulla. The ampulla divides into a short oviduct, which enters the female gland mass and the more elongate vas deferens. The proximal portion of the vas deferens widens into a wide, highly digitate, flattened prostatic portion, which envelops most of the bursa copulatrix. From the distal end of the prostatic portion the vas deferens narrows abruptly into the elongate, convoluted ejaculatory portion which is highly muscular. The ejaculatory portion of the vas deferens remains relatively narrow throughout its length and does not widen at the penial bulb, which

is relatively short. The penis contains approximately six rows of curved, acutely pointed penial hooks which are approximately 10–15 μm wide at the base and 15–20 μm in length (Figure 3C–E). The vaginal opening is enlarged and muscular. The vagina is relatively narrow and straight. At its proximal end is a large, thin-walled, spherical bursa copulatrix. The slender receptaculum seminis is extremely elongate and curved. The duct of the receptaculum seminis joins the vagina distally, near the base of the vaginal duct. From the middle of the receptaculum duct, the uterine duct emerges and joins the female gland mass. The female gland mass is large and completely developed in all specimens examined.

Developmental biology: Egg ribbons (Figure 4B) are flat, transparent spirals consisting of three whorls and containing yellow eggs. A 15 mm adult produced an 8 mm ribbon with 2000 eggs. Each capsule has one egg, 240 μm wide. The larvae initially almost fill this capsule. Eggs can vary from 120 to 300 μm wide. The occurrence of two larvae per egg is rare. Lecithotrophic larvae hatch after 31 days at 14°C, with type B shell shape (Todd, 1981) previously known as type I (Thompson, 1961). Some species of opisthobranchs exhibit poecilogony and can change their developmental strategy from lecithotrophic larvae or direct development to planktotrophic larvae in response to adult starvation or other environmental variation (Clark & Goetzfried, 1978). Animals from the Hill Street population were subjected to starvation but did not demonstrate any change in reproductive strategy.

Population genetics: The genetic identity measure, *I* was 0.374 between the species *Doriopsilla albopunctata* and *Doriopsilla gemela*. Both species were collected from Hill Street, San Diego County for the interspecific allozyme comparison. Typically, populations of congeneric species have *I* values from 0.3 to 0.8 (Thorpe, 1983; Nei, 1987). The genetic distance value, *D*, was 0.983 between these two species. Allelic frequencies were within expected parameters as to indicate Hardy-Weinberg Equilibrium, as determined by chi square analysis, with the exception of MDH, which occurred as three homozygotic alleles with no detected heterozygotes. This is significant at the 0.005 level, as determined by chi square analysis. Details of methodology and banding patterns are presented by Schaefer (in preparation).

DISCUSSION

The systematics of the genera within the Dendrodorididae has been historically the subject of considerable confusion. The names *Dendrodoris* Ehrenberg, 1831, *Doriopsis* Pease, 1860, *Doridopsis* Alder & Hancock, 1864, and *Doriopsilla* Bergh, 1880, have been applied to various members of the family. Pruvot-Fol (1930) showed that *Doridopsis* is a junior synonym of *Dendrodoris*, but con-

sidered *Doriopsis* to be a member of the Archidorididae. Subsequently, *Doriopsis* has been placed in the Dorididae (Kay & Young, 1969). Regardless of its familial placement, *Doriopsis* has a well-developed radula and pectinate gills and is clearly not a dendrodorid.

Eliot (1906) recognized the distinction between *Dendrodoris* (as *Doridopsis*) and *Doriopsilla*, with the former having elongate buccal nerves and more posteriorly situated buccal ganglia. Valdés & Ortea (1997:240) questioned this distinction and stated that Eliot was incorrect in stating that the buccal ganglia in *Doriopsilla* were situated anteriorly. However, they did not describe or illustrate the position of the buccal ganglia of any of the species they described. The differences in buccal ganglion position noted by Eliot has been confirmed by other authors. Marcus (1957:fig.) and Edmunds (1971:figs. 21d, 22d) have shown the more posterior position of the ganglia in several species of *Dendrodoris*. Several other authors (Marcus, 1961:fig. 19; Marcus & Marcus, 1967:fig. 62; Gosliner, 1991:fig. 8) have indicated that the position of the buccal ganglia in species of *Doriopsilla* is within the anterior nerve ring. The two species of *Doriopsilla* examined here also have the buccal ganglia situated in the nerve ring rather than more posteriorly. Most subsequent workers have considered the two genera as distinct with the exception of Thompson (1975:500) who stated that "the distinction is based upon several features of the morphology which appear to me inadequate," but provided no further details. Most recently, Valdés (1996) provided phylogenetic evidence that species of *Doriopsilla* and *Dendrodoris* form monophyletic sister clades. The present species possess the synapomorphic features of an eccentric anus, a flattened prostate, and penial spines with an elongate base, which characterize members of *Doriopsilla* (Valdés & Ortea, 1997).

Systematic confusion has surrounded the systematics of the Dendrodorididae from the Pacific coast of North America. Cooper (1863) described *Doris albopunctata* from Santa Barbara, California. Only the external coloration was described and no type material is extant (MacFarland, 1905). MacFarland (1905) later described *Doriopsis fulva* from Monterey Bay, California. Similarly, he described only the external anatomy and coloration of the living animal. In his original description, MacFarland noted that *D. fulva* was possibly identical with Cooper's species, but stated it was difficult to be certain, based on the superficial description and the absence of Cooper's type material. Subsequently (1906), he described and illustrated aspects of the reproductive system of *D. fulva*. Cockerell & Eliot (1905) described *Doridopsis reticulata* from San Pedro, California. They described the external morphology and a few aspects of the internal anatomy. They also stated that their species might be identical to *Doris albopunctata* Cooper. Steinberg (1961) considered these three species as synonymous, the differences being based largely on a darker coloration of spec-

imens from southern California. Roller (1970) and McDonald (1983) also considered these three names as synonyms. Behrens (1980, 1991) considered *Dendrodoris fulva* as distinct from *Doriopsilla albopunctata*. Our examination of specimens of *Doriopsilla albopunctata* from Baja California to northern California revealed considerable variation in the body color, similar to that described by Steinberg (1961). Specimens from northern California are light yellow throughout, but may occasionally have a central patch of chestnut brown on the dorsum. Specimens from southern California exhibit much more variation in color than do northern California specimens, but are generally much darker in color. There were no other significant anatomical differences between specimens of different color forms or from different localities. All specimens with white gills produced egg masses with a spiral attached to the substrate by its inner edge. All of these egg masses yielded planktotrophic larvae. Analysis of allozyme frequencies yielded no significant differences within or between widely separated populations. Analysis of morphological, developmental and genetic data support the conclusions of Steinberg (1961), Roller (1970), and McDonald (1983), that *Doris albopunctata* Cooper, 1863, *Doriopsis fulva* MacFarland, 1905, and *Doridopsis reticulata* Cockerell & Eliot, 1905, represent a single species with Cooper's name having priority as the senior synonym. There is no doubt that the material described by Cockerell & Eliot (1905) and MacFarland (1906) are conspecific with the present material. Cockerell & Eliot described an elongate receptaculum seminis (as "spermatocyst") as in the present material. MacFarland (1906:fig. 38) illustrated part of the reproductive system of *Doriopsis fulva* with a receptaculum seminis (as "spermatocyst") which exceeds the length of the bursa copulatrix (as "spermatotheca") and which enters the vagina proximally, as in the present material.

Material that Behrens (1980, 1991), McDonald & Nybakken (1980), and McDonald (1983) considered as a distinct species is conspecific with *Doriopsilla gemela*.

Doriopsilla gemela is morphologically distinct from *D. albopunctata*. Externally, specimens of *D. gemela* have deep yellow or yellow-orange gills, whereas those of *D. albopunctata* are white or pale yellow. There are fewer rhinophoral lamellae (7–10) in *D. gemela* than in *D. albopunctata* (11–30). The larger tubercles of *D. albopunctata* contain glands, whereas the smaller ones of *D. gemela* do not. The remainder of the external anatomy is extremely similar between the two species.

The internal anatomy of the two species differs consistently in many significant regards. The glandular portion of the esophagus of *D. gemela* is more elongate and convoluted than in *D. albopunctata*. More posteriorly, *D. gemela* has an elongate muscular portion of the esophagus, while that of *D. albopunctata* is short. The digestive gland lobes are well separated and lobed on the outer edges in *D. gemela*, while they are partially fused in *D.*

albopunctata. The intestinal caecum is readily visible in *D. gemela*, but is more ventral and obscured by the glandular portion of the intestinal lobes in *D. albopunctata*.

There are also consistent differences in the reproductive anatomy of the two species. The ejaculatory portion of the vas deferens of *D. gemela* is elongate and consists of many convolutions, whereas in *D. albopunctata* the ejaculatory segment is shorter and thicker and without convolutions. In *D. gemela* there are about six rows of small penial spines that are 15–20 µm in length. In contrast, in *D. albopunctata* there are about 16 rows of spines that are about 50 µm in length. The receptaculum seminis of both species is fairly elongate, but in *D. albopunctata* it is more elongate and extends beyond the proximal extreme of the bursa copulatrix. More significantly, the duct of the receptaculum seminis of *D. gemela* enters the distal portion of the vagina, while in *D. albopunctata* it enters the proximal extreme, near the base of the bursa copulatrix.

The two species differ markedly in their developmental biology. In *D. gemela* the egg ribbon is flat against the substrate, while in *D. albopunctata* it is attached by its narrow edge and well elevated from the substrate. In *D. gemela* the yolky eggs develop into lecithotrophic larvae, while the larvae of *D. albopunctata* are planktotrophic. Genetic distances in allozyme frequencies are also consistent with distinct congeners.

Among described species of *Doriopsilla*, *D. gemela* is unique in having the receptaculum duct enter the vaginal duct basally. The presence of an elongate muscular portion of the esophagus is identical to that described for *D. rowena* Marcus & Marcus, 1967 (Marcus & Marcus, 1967:206, fig. 62b), rather than a short muscular portion described here for *D. albopunctata* and previously for *D. janaina* Marcus & Marcus, 1967 (Gosliner, 1991:292, fig. 8). These differences in digestive anatomy between *D. gemela* and *D. albopunctata* suggest that despite their similarity in external morphology and coloration, they are both anatomically more similar to other species than to each other. Morphological data from other species of *Doriopsilla* is necessary to develop hypotheses of phylogeny and test the suggestion that *D. gemela* is more closely related to *D. rowena* than to *D. albopunctata* and *D. janaina*.

Valdés & Ortea (1997) suggested that *Doriopsilla rowena* and *D. janaina* might be possible synonyms of *D. areolata* Bergh, 1880. However, *D. janaina* differs from *D. areolata* in having a short rather than elongate muscular portion of the esophagus (Gosliner, 1991). Based on the description of Marcus & Marcus (1967:fig. 62c), *D. rowana* differs from *D. areolata* in that the duct of the receptaculum seminis is separated from the bursa copulatrix, whereas in *D. areolata* it enters directly at the base of the bursa (Valdés & Ortea, 1997:fig. 4b). The color patterns of these species also differ from the variation described for any of the subspecies of *D. areolata*. It

would appear that *D. rowena* and *D. janaina* represent distinct species.

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LITERATURE CITED

- BEHRENS, D. W. 1980. Pacific Coast Nudibranchs, a Guide to the Opisthobranchs of the Northeastern Pacific. Sea Challengers: Los Osos, California. 112 pp.
- BEHRENS, D. W. 1991. Pacific Coast Nudibranchs, a Guide to the Opisthobranchs. Alaska to Baja California. 2nd ed. Sea Challengers: Monterey, California, 107 pp.
- CLARK, K. B. & A. GOETZFRIED. 1978. Zoogeographic influences on development patterns of North Atlantic Ascoglossa and Nudibranchia, with a discussion of factors affecting egg size and number. *Journal of Molluscan Studies* 44(3):283–294.
- COCKERELL, T. D. A. & C. ELIOT. 1905. Notes on a collection of Californian nudibranchs. *Journal of Malacology* 12(3):31–53.
- COOPER, J. G. 1863. On new or rare Mollusca inhabiting the coast of California. 2. Proceedings of the California Academy of Natural Sciences 3:56–60.
- EDMUNDS, M. 1971. Opisthobranchiate Mollusca from Tanzania (Suborder: Doridacea). *Zoological Journal of the Linnean Society* 50(4):339–396.
- ELIOT, C. N. E. 1906. The genus *Doriopsilla* Bergh. *Journal of Conchology* 11(12):366–367.
- GOSLINER, T. M. 1991. The opisthobranch gastropod fauna of the Galapagos Islands. Pp. 281–305 in M. James (ed.), *Galapagos Marine Invertebrates*. Plenum: New York.
- HAVENHAND, J. N., J. P. THORPE & C. D. TODD. 1986. Estimates of biochemical genetic diversity within and between the nudibranch molluscs *Adalaria proxima* (Alder and Hancock) and *Onchidoris muricata* (Doridacea: Onchidorididae). *Journal of Experimental Marine Biology and Ecology* 95:105–112.
- KAY, E. A. & D. K. YOUNG. 1969. The Doridacea (Opisthobranchia; Mollusca) of the Hawaiian Islands. *Pacific Science* 23(2):172–231.
- MACFARLAND, F. M. 1905. A preliminary account of the Dorididae of Monterey Bay, California. Proceedings of the Biological Society of Washington 18:35–54.
- MACFARLAND, F. M. 1906. Opisthobranchiate Mollusca from Monterey Bay, California, and vicinity. *Bulletin of the United States Bureau of Fisheries, Washington, D.C.* 25:109–151.
- MARCUS, ER. 1957. On Opisthobranchia from Brazil 2. *Journal of the Linnean Society of London*, 43:390–486.

- MARCUS, ER. 1961. Opisthobranchia from North Carolina. The Journal of the Elisha Mitchell Scientific Society 77(2):141-151.
- MARCUS, ER. & EV. MARCUS. 1967. American Opisthobranch Mollusks. University of Miami Institute of Marine Sciences: Miami. 256 pp.
- MCDONALD, G. 1983. A review of the nudibranchs of the California coast. Malacologia 24(1-2):114-276.
- MCDONALD, G. & J. NYBAKKEN. 1980. Guide to the Nudibranchs of California. American Malacologists: Melbourne, Florida. 72 pp.
- MORROW, C. C., J. P. THORPE & B. E. PICTON. 1992. Genetic divergence and cryptic speciation in two morphs of the common subtidal nudibranch *Doto coronata* (Opisthobranchia: Dendronotacea: Dotoidae) from the northern Irish Sea. Marine Ecology Progress Series 84:53-61.
- NEI, M. 1972. Genetic distance between populations. American Naturalist 106:283-292.
- NEI, M. 1987. Molecular Evolutionary Genetics. Columbia University Press: New York. 512 pp.
- ROLLER, R. A. 1970. A list of recommended nomenclatural changes for MacFarland's "Studies of opisthobranchiate mollusks of the Pacific coast of North America". The Veliger 12(3):371-374.
- PRUVOT-FOL, A. 1930. Du genre *Dendrodoris* Ehrenberg et de ses rapports avec le genre *Doriopsis* Pease et avec quelques autres. Note sur la taxonomie des nudibranches. Bulletin du Museum d'Histoire Naturelle, Paris (2)2(3):291-297.
- STEINBERG, J. E. 1961. Notes on the opisthobranchs of the West Coast of North America. 1. Nomenclatorial changes in the order Nudibranchia (Southern California). The Veliger 4(2): 57-63.
- THOMPSON, T. E. 1961. The importance of the larval shell in the classification of the Sacoglossa and the Acoela (Gastropoda: Opisthobranchia). Proceedings of the Malacological Society of London 34(5):233-238.
- THOMPSON, T. E. 1975. Dorid nudibranchs from eastern Australia (Gastropoda, Opisthobranchia). Journal of Zoology 176(4): 477-517.
- THORPE, J. P. 1983. Enzyme variation, genetic distance and evolutionary divergence in relation to levels of taxonomic separation. Pp. 131-152 in G. S. Oxford & D. Rollinson (eds.), Protein Polymorphism: Adaptive and Taxonomic Significance. Academic Press: London.
- TODD, C. D. 1981. Ecology of nudibranch molluscs. Oceanographic Marine Biology 19:141-234.
- TODD, C. D. 1983. Reproductive and trophic ecology of nudibranch molluscs. Pp. 225-255 in W. D. Russell-Hunter (ed.), The Mollusca. Ecology. Vol. 6. Academic Press: New York.
- VALDÉS, A. 1996. Revisión de la superfamilia Porodoridoidea Odhner en Franc, 1968 (Mollusca: Nudibranchia) en el Océano Atlántico. Ph.D. thesis, Universidad de Oviedo.
- VALDÉS, A. & J. ORTEA. 1997. Review of the genus *Doriopsis* Bergh, 1880 (Gastropoda: Nudibranchia) in the Atlantic Ocean. The Veliger 40(3):240-254.